Journal of Pure and Applied Agriculture (2020) 5(4): 34-41
ISSN (Print) 2617-8672, ISSN (Online) 2617-8680
http://jpaa.aiou.edu.pk/

RESEARCH PAPER

Orchard locality and postharvest oxalic acid application influence fruit quality and shelf life of Jamun (*Syzygium cumini* L.) fruit

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Received: 12 October 2020
Accepted: 23 December 2020

Key Message: Effect of orchard location and postharvest oxalic acid (OA) treatment on fruit quality and shelf life of jamun was evaluated. Postharvest dip of 2mM-OA revealed better fruit quality and shelf life. Moreover, harvest locations have a significant impact on fruit quality.

Abstract: Jamun is a highly nutritious fruit crop having a short shelf life. This study was performed to evaluate the effect of harvest location and oxalic acid treatment on fruit quality, shelf life, biochemical attributes and antioxidative activity in jamun fruit under ambient conditions. The fruit was sourced from two different locations in the Multan region and subjected to various postharvest oxalic acid (OA) applications (T0: Control, T1: 0.5 mM, T2: 1 mM, T3: 2 mM). The treated fruits were subjected to physico-chemical evaluation at shelf under ambient environment (25±2 ºC & 55-60% RH). Harvest location significantly affected various physical, biochemical and antioxidative attributes of jamun fruit. The fruit harvested from Orchard-II exhibited lower weight loss and fruit shriveling with higher titratable acidity (TA), vitamin C, total phenolic contents (TPC) and enzyme activity of superoxide dismutase (SOD), while, soluble solid contents (SSC), SSC: TA ratio, total antioxidants and activities of catalase (CAT) and peroxidase (POX) enzymes was observed in Orchard-I. Irrespective of orchard locality and shelf life, the jamun fruit treated with higher dose of OA (2 mM-OA) exhibited significant lower fruit weight loss, shriveling % with better fruit biochemical and antioxidative attributes as compared to untreated fruit and other treatments. Fruit weight loss, shriveling percentage, SSC, SSC: TA ratio showed a significant linear increasing trend, however TA, vitamin C, total antioxidants and enzymatic activity of SOD and CAT of jamun fruit significantly decreased with the shelf life. Conclusively, the fruit harvested from orchard–II and higher dose of OA jamun fruit revealed better fruit quality during shelf due to lower weight loss and shriveling by increasing antioxidant system of the fruit.

Keywords: Anti-oxidative enzymes, Harvest location, Phytochemicals, Postharvest, *Syzygium cumini*

Abbreviations: OA = Oxalic acid; SSC = Soluble solid contents; TA = Titratable acidity; TPC = Total phenolic contents; SOD = Superoxide dismutase; CAT = Catalase; POX = Peroxidase

To cite this article: Aslam, M. M., Ullah, S., Razzaq, K., Rajwana, I. A., Akhtar, G., Faried, H. N., Amin, M., Naeem-Ullah, U., & Khalil, U. (2020). Orchard locality and postharvest oxalic acid application influence fruit quality and shelf life of Jamun (*Syzygium cumini* L.) fruit. *Journal of Pure and Applied Agriculture, 5*(4), 34-41.

Introduction

A wide range of fruit crops has been grown in Punjab province of Pakistan including some major and minor fruits. There is a need to explore the potential of some minor fruits that might contribute to the economy of the country under current economic crises and challenges faced by the agricultural sector due to climate change. Jamun (*Syzygium cumini* L.) belongs to the family *Myrtaceae,* and grown in both tropical and subtropical climatic regions. It is a minor fruit of Pakistan, mostly jamun is cultivated on the main boundaries of fruit orchards and fields as shelter belts in the country while few commercial orchards have been reported in Faisalabad region of Punjab (Din et al., 2020). However, it is very popular among the fruit growers owing to its high nutritive, medicinal value and wide range of application in the processing industry. Currently, jamun fruit production is facing different challenges including lack of elite cultivars, poor quality, higher fruit losses, disease incidence, short shelf life and improper packaging.

Various factors have been reported to influence the fruit quality including geographic location of cultivation (Dragovic-Uzelac et al., 2007). Different orchards locations have different environmental factors including light, temperature and moisture content that have major roles in both on-tree and postharvest fruit quality (Ullah et al., 2013). Previously, fruit physical, biochemical and sensory attributes have been reported in mango, pear, apricot (Luton & Holland, 1986; Dragovic-Uzelac et al., 2007; Khan et al., 2009). In jamun fruit, Din et al. (2020) found a huge variability especially in its biochemical attributes like SSC, TA and phenolic contents when different accessions were harvested from various cities of Punjab province.

Oxalic acid (OA) is present in various fruits with variable concentrations. Various biochemical attributes are influenced by OA application (Shimada et al., 1997). Postharvest treatments of OA have been reported to play a vital role in fruits for retaining fruit quality attributes and shelf life by delaying the ripening process (Zheng et al., 2012) as well as fruit senescence (Wu et al., 2011). It acts as an anti-oxidative agent by scavenging the free radical produced during fruit ripening (Zheng et al., 2007). Moreover, application of OA has reported to reduce the postharvest diseases development in jujube fruit (Wang et al., 2009). Previously postharvest treatments of OA have been reported to delay fruit senescence, reduce fruit firmness losses, reduce anthocyanin and reddening enzyme, increase disease resistance and chilling tolerance in jujube, plum, kiwi, mango and peach fruit (Wang et al., 2009; Wu et al., 2011; Huang et al., 2013; Razzaq et al., 2015; Zhu et al., 2016; Razavi & Hajilou, 2016). Moreover, OA treatment, applied before harvesting, has also been reported in improving plant yield and fruit physical quality parameters like firmness and color in pomegranate (García-Pastor et al., 2020).

During postharvest life, fruits face various physical stresses that result in higher production of reactive oxygen species (ROS) which can cause damage to cellular organelles (Pakkish et al., 2019). Fruits adopt various defense mechanisms by producing different bioactive compounds and antioxidative enzymes that can neutralize the ROS (Madani et al., 2019). To understand the mechanism for retention of fruit quality during shelf life, it is important to study the level of such bioactive compounds- like phenolic contents and total antioxidants and activity of antioxidative enzymes. To the best of our knowledge, no information exists in literature on the role of oxalic acid harvested from different locations on fruit physical, biochemical and anti-oxidative attributes of jamun fruit at shelf after harvest. So the impact of orchard location on the quality of jamun fruit need to be explored under the climatic condition of Punjab, Pakistan. The change in climatic conditions of locality and above beneficial effects of oxalic acid, it is hypothesized that the oxalic acid will improve quality and shelf life of jamun fruit harvested from different orchards. Therefore, this study was planned to evaluate the OA effect on the quality and shelf life of jamun fruit harvested from different locations.

Materials and Methods

Fruit harvesting

The healthy and blemished free fruit was harvested at commercial maturity from two different orchards located in Multan. Orchard-I was located Near Laeeq Rafique Hospital, Bahawalpur Road, Multan (71°49′ 75″ E; 30°03′95″ N) while Orchard-II was located at Butch Villas, Bosan Road, Multan (71°49′ 58″ E; 30°28′58″ N). The harvested fruit were dipped in oxalic acid (OA) treatments (T0: Control, T1: 0.5m*M*, T2: 1m*M*, T3: 2 m*M*) for 10 minutes using 0.01% Tween 20® as a surfactant. After application, the treated fruit was air dried in shade and packed in cardboard boxes. The boxes were transported to Postharvest Science and Technology Lab, MNS University of Agriculture Multan and kept at room temperature for the shelf life and quality assessment under ambient conditions (25 ± 2 °C; 55-60% R.H.). The experiment was conducted under CRD factorial arrangement including harvest location, OA-treatment and shelf period as factor with three replications.

Physical quality of fresh jamun fruit

Jamun fruits were kept on the shelf for the assessment of weight loss. It was calculated by using the formula described by Amin (2012). Fruit weight of each sample was recorded daily using weight balance and weight loss (%) was measured according to following equation:

$$\text{Physical fruit weight loss (\%)} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100$$

Twenty five fruits were kept at shelf for the assessment of shriveling by using the scale ranging from 1 to 4 (0-25% shriveling) to (75-100% shriveling), respectively as described by Amin (2012) and average was expressed in percentage of fruit shriveling.

Biochemical estimation of fresh jamun fruit

Soluble solid contents (SSC) were determined by using a hand digital refractometer (PAL-1, Atago, Tokyo, Japan) and was expressed as °Brix. Titratable acidity (TA) was calculated by titraing juice of jamun fruit against 0.1 *N* NaOH, the method described by Ali et al. (2011) and given in percentage. Ripening index (SSC: TA ratio) was determined by dividing SSC with respective TA (titratable acidity) of fruits. Vitamin C was estimated by using protocol as explained by the Ullah et al. (2013).

Total antioxidant and total phenolic contents

Total antioxidant capacity was determined using the 2, 2-diphenyl-1-picrylhydrazyl assay (DPPH) as proposed by Mimica-Dukic et al. (2003). The TPC of jamun fruit was measured following the method of Ainsworth & Gillespie (2007) and gallic acid was used as standard with some modifications. Absorbance was recorded to determine total antioxidants and TPC using spectrophotometer (Cecil Aquarius, CE 7400S, Cecil Instruments, U.K.) at 520 and 760 nm, respectively.

Determination of antioxidative enzymes activities

The superoxide dismutase (SOD) activity was determined by measuring the 50% inhibition of the photochemical reduction of nitro blue tetrazolium (NBT) according to method adopted by Stajner & Popovic (2009) and absorbance was recorded at 560 nm. One unit of SOD activity was defined as "the amount of enzyme that inhibited 50% of NBT photoreduction as expressed (U mg $protein^{-1}$). Catalase (CAT) enzyme activity was determined by the protocol adopted by Ullah et al. (2013) and taking absorbance at 240nm using spectrophotometer. The activity of POX was determined by adopting the method of Liu et al. (2009) and absorbance was recorded at 470 nm. The 1 unit of POX activity was defined as "an absorbance change in 0.01 units per min". POX activity was calculated and expressed as U mg^{-1} protein.

Statistical analysis

Statistical analysis was done at five percent level of significance by using statistix®8.1 software. Mean significance was determined by using the Fisher's Least Significant Difference (LSD) test (Steel & Torrie., 1997).

Results

Weight loss and fruit shriveling

Jamun fruits harvested from different locations exhibited significantly ($P \leq 0.05$) different weight loss irrespective to OA treatments. The fruit harvested from the orchard-I showed 10% more weight loss than orchard-II (Fig. 1A). Oxalic acid treatments significantly reduced the fruit weight loss and a 15% lower weight loss was recorded in 2 m*M*-OA treated fruit as compared to control (Fig. 1C). During ambient storage of jamun, the weight loss increased significantly with linear trend at shelf from day-1 to day-4 of storage at shelf. It was recorded maximum at last day viz., day-4 (Fig. 1E). Harvest location significantly affected the fruit shriveling percentage ($P \leq 0.05$) regardless of OA-treatments and days at shelf. Significantly lower fruit shriveling (28%) was recorded in fruits harvested from orchard-II as compared to orchard- I (Fig. 1B). The fruit treated with postharvest oxalic acid showed significantly lower shriveling percentage than untreated fruit harvested from both the locations during storage at ambient conditions. About 19% lower shriveling percentage was recorded as a result of 2 m*M*-OA application when compared to untreated fruit (Fig. 1D). Shriveling increased with respect to days during ambient storage in significant linear trend irrespective to harvest location and OA-treatments (Fig. 1F).

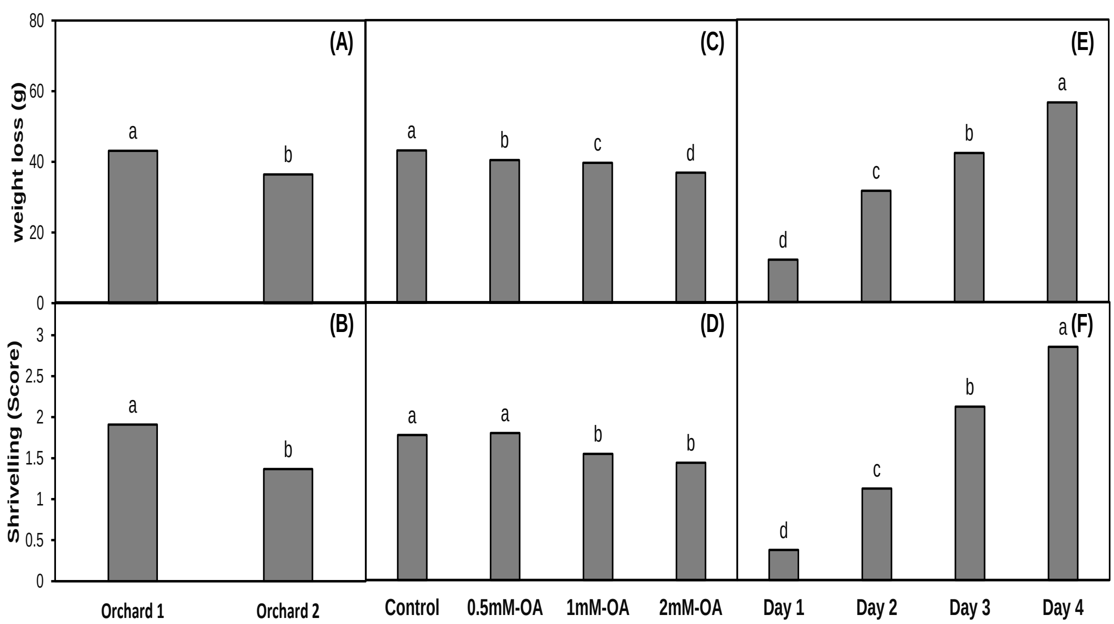

Fig.1 Fruit weight loss and shriveling of jamun fruit as affected by harvest location (A,B), postharvest oxalic acid treatments (C,D) at shelf (E,F) under ambient conditions. The mean not sharing same letter differ significantly from each other at $P \leq 0.05$; $n_{(harvest\ location)} = 48$; $n_{(oxalic\ acid\ treatments)} = 24$; $n_{(days\ at\ shelf)} = 24$

Biochemical attributes

SSC, TA and SSC/TA

Irrespective to OA-treatment and days at shelf, jamun fruit SSC were significantly ($P \leq 0.05$) affected by harvest location (Fig. 2A). When averaged the mean values, the fruit harvested from orchard-I exhibited about 25% higher SSC than the fruit harvested from Orchard-II. Postharvest oxalic acid treatment showed significant lower SSC than untreated fruit harvested from both the locations during storage at ambient conditions (Fig. 2D). The fruit treated with 2 m*M*-OA exhibited about 15% lower SSC than controlled fruit. The SSC of jamun fruit increased with increase in shelf period upto day-3 irrespective to harvest location and OA-treatment (Fig. 2G). Jamun fruits harvested from different locations were significantly different for titratable acidity percentage irrespective to OA treatments and shelf intervals (Fig. 2B). Regarding the effect of OA treatment, TA in 2 *mM*-OA treated jamun fruits was 4% lower than control fruits (Fig. 2E). During shelf life, slight increase in TA was recorded that peaked over the last day of ambient storage (Fig. 2H). The SSC/TA ratio irrespective to OA treatments and shelf intervals were significantly affected by fruit harvest location ($P \leq 0.05$). The ripening index (SSC/TA) was 16.41% higher in jamun fruits harvested from orchard-II (Fig. 2C). The fruit treated with postharvest oxalic acid showed a significantly lower SSC/TA ratio than untreated fruit harvested from both the locations during storage at ambient conditions. The fruits treated with 0.5 m*M*-OA exhibited 12% lower SSCTA ratio than untreated jamun fruits (Fig. 2F). Irrespective to OA treatments and harvest location SSC/TA significantly increased ($P \leq 0.05$) at various days of shelf storage (Fig. 2I).

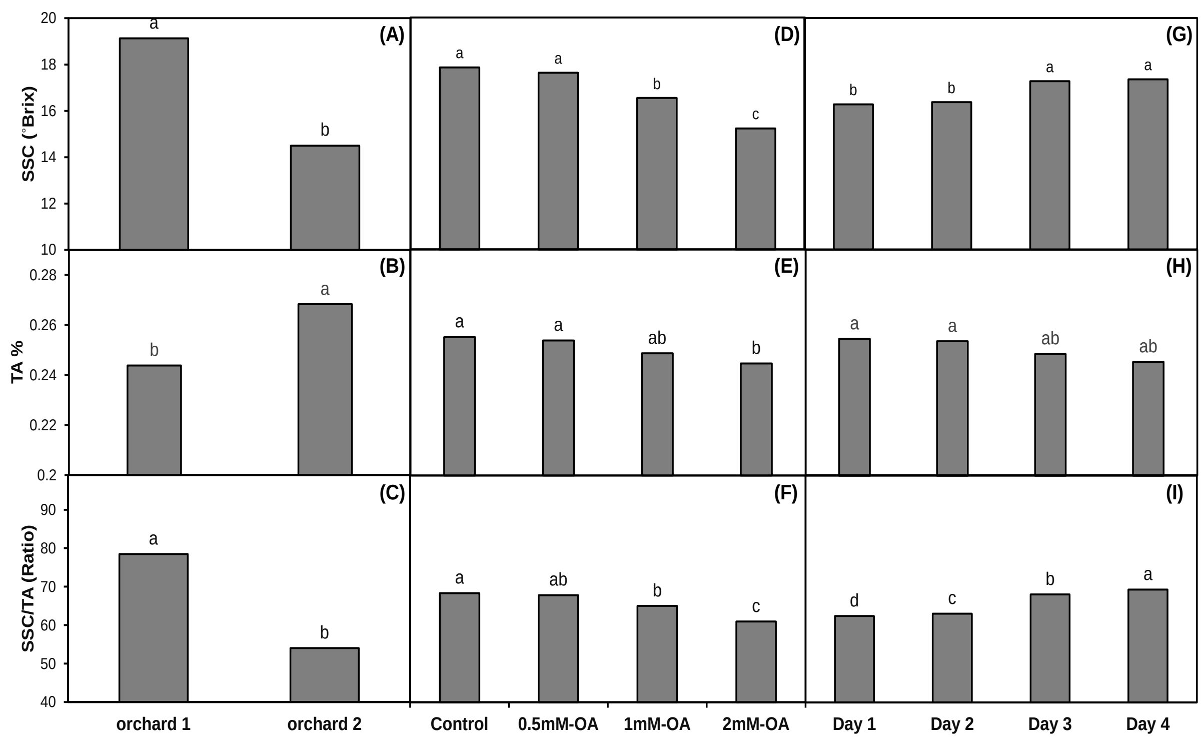

Fig. 2 SSC, TA and SSC/TA of jamun fruit as affected by harvest location (A-C), postharvest oxalic acid treatments (D-F) at shelf (G-I) under ambient conditions. The mean not sharing same letter differ significantly from each other at $P \leq 0.05$; $n_{(\text{harvest location})} = 48$; $n_{(\text{oxalic acid treatments})} = 24$; $n_{(\text{days at shelf})} = 24$

Vitamin C, TPC and total antioxidants

The vitamin C was found significantly different ($P \leq 0.05$) for fruit harvested from both locations irrespective to OA treatments and shelf intervals. The jamun fruits harvested from orchard-II exhibited 58% higher vitamin c contents than orchard-I (Fig 3A). Postharvest application of OA-significantly retains the vitamin c as compared to the control fruit in both locations through fruit ripening. The OA-treated fruits were higher in vitamin C contents than untreated jamun fruits. The fruits treated with 2 m*M*-OA exhibited 58% more vitamin c then untreated jamun fruits (Fig. 3D). The vitamin C increased at shelf irrespective to harvest location and OA treatments (Fig. 3G). The harvest location exhibited a significant effect ($P \leq 0.05$) upon jamun fruit TPC irrespective of OA treatments and days at shelf. The jamun fruits harvested from orchard-II had 79% more TPC than orchard-I (Fig. 3B). The OA-treated fruits were higher in TPC contents than untreated jamun fruits. The fruits treated with 1 m*M*-OA exhibited 5% more TPC than untreated jamun fruits (Fig. 3E). It was non-significantly different at various shelf days (Fig. 3H). Irrespective of OA-treatment and days at shelf, jamun fruit total antioxidants were significantly affected by harvest location. The fruits harvested from orchard-I were 19% higher in total antioxidant activity than fruit harvested from orchard-II (Fig. 3C). The OA-treated fruits were higher in total antioxidant than untreated jamun fruits. The fruits treated with 2 m*M*-OA exhibited 13% more total antioxidants than untreated jamun fruits (Fig. 3F). It was significantly increased ($P \leq 0.05$) at various shelf days (Fig. 3I).

Antioxidative enzymes (SOD, CAT & POX)

SOD enzyme activity was significantly affected ($P \leq 0.05$) by harvest location irrespective to OA treatments and storage days (Fig 4A). The fruits harvested from orchard-II were 25% higher in SOD activity than those of orchard-I. The SOD activity was significantly lower in OA treated fruits than control fruit ($P \leq 0.05$). The fruits treated with 2m*M*-OA exhibited 21% less SOD activity than control (Fig. 4D). SOD activity was significantly decreased ($P \leq 0.05$) in linear trend when kept at shelf for four days (Fig. 4G). The catalase (CAT) enzyme activity was significantly ($P \leq 0.05$) affected by harvest location regardless of OA-treatments and days at shelf. The CAT activity was 31% higher in fruits harvested from orchard-I in comparison to orchard-II (Fig. 4B). As far as OA treatment is concerned, the jamun fruits treated with 2 m*M*-OA exhibited 43% higher CAT activity than control (Fig. 4E). A significant ($P \leq 0.05$) linear decrease in CAT enzyme activity was recorded during shelf life (from 0 to 4th day) (Fig. 4H). Harvest location, irrespective to OA treatments and days at shelf, significantly affected ($P \leq 0.05$) the POX activity of jamun fruits which was 20 % higher in fruits harvested from orchard-I as compared to orchard-II (Fig. 4C). OA increased overall POX activity of jamun and maximum activity was recorded as a result of 2m*M*-OA treatment that was 27% higher than control (Fig. 4F). POX activity increased from day-1 to day and remained constant up to day-4 when kept at ambient for shelf life evaluation (Fig. 4I).

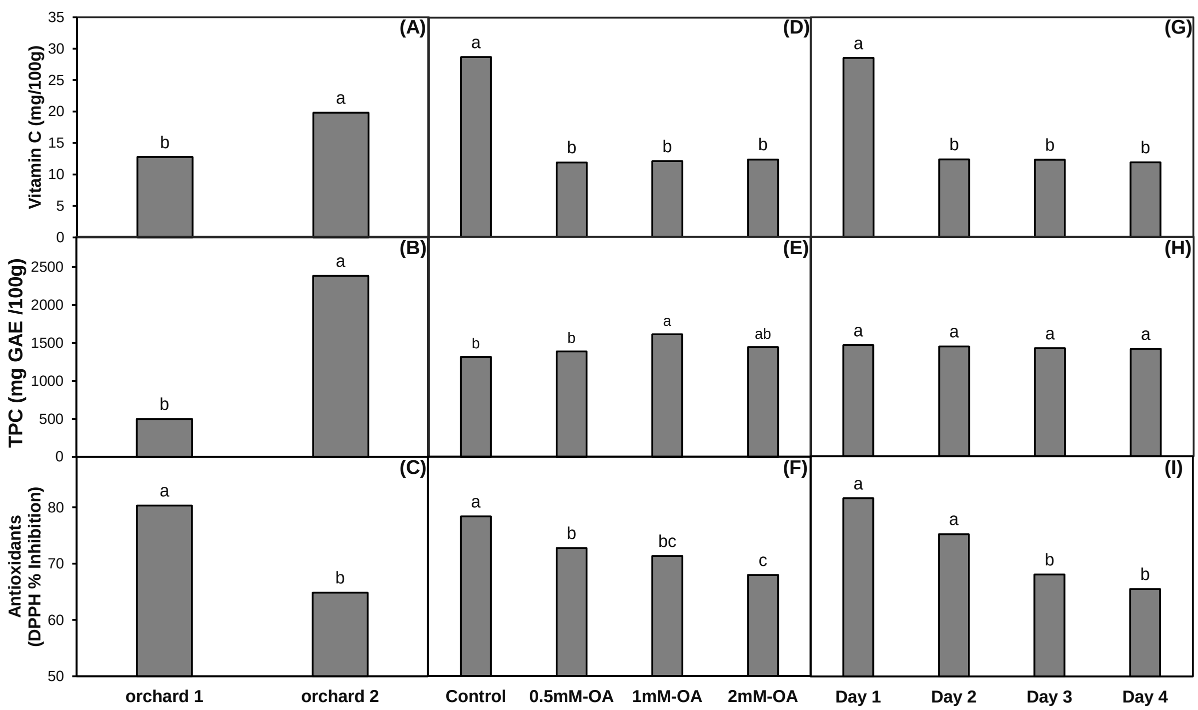

Fig. 3 Vitamin C, Total Phenolic Contents and antioxidant capacity of jamun fruit as affected by harvest location (A-C), postharvest oxalic acid treatments (D-F) at shelf (G-I) under ambient conditions. The mean not sharing same letter differ significantly from each other at $P \leq 0.05$; $n_{(harvest\ location)} = 48$; $n_{(oxalic\ acid\ treatments)} = 24$, $n_{(days\ at\ shelf)} = 24$

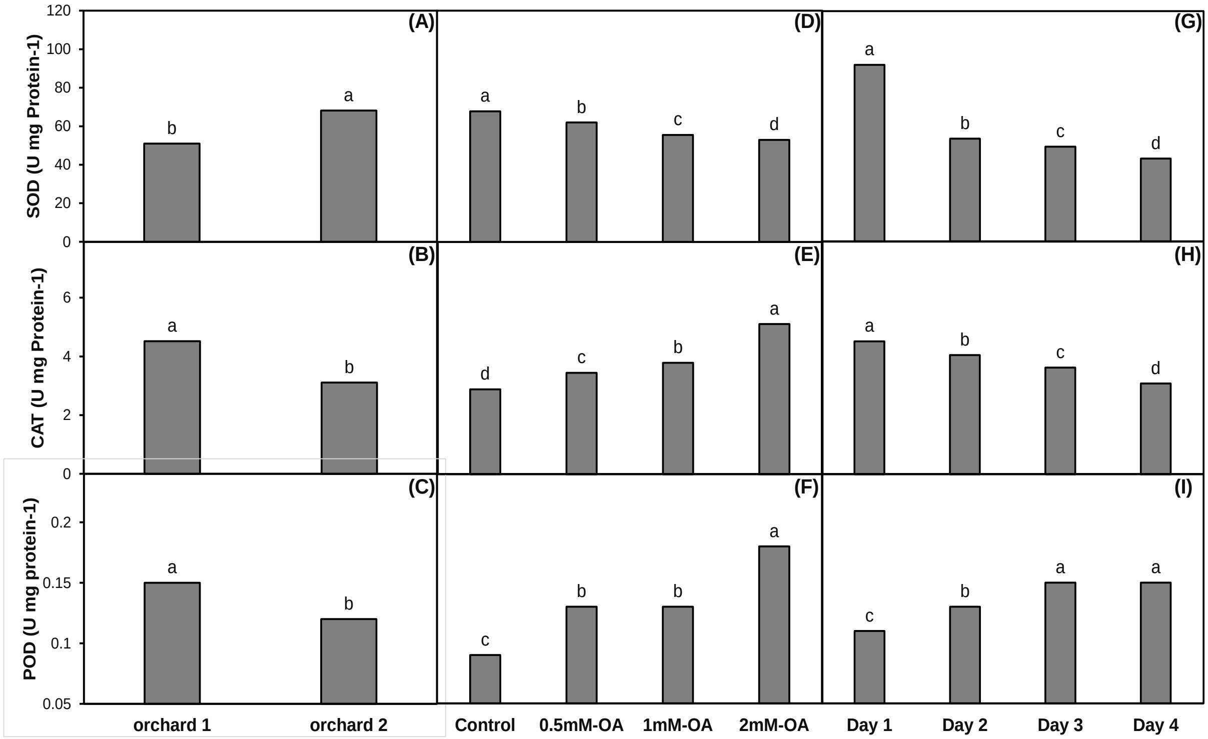

Fig. 4 SOD, CAT and POD enzyme activities of jamun fruit as affected by harvest location (A-C), postharvest oxalic acid treatments (D-F) at shelf (G-I) under ambient conditions. The mean not sharing same letter differ significantly from each other at $P \leq 0.05$; n($_{\text{harvest location}}$) = 48; n($_{\text{oxalic acid treatments}}$) = 24; n($_{\text{days at shelf}}$) = 24

Discussion

In current study, weight loss and fruit shriveling percentage was significantly lower in fruit harvest from orchard-II as compared to orchard-I. Fruit weight loss not only depends upon the postharvest condition but various pre-harvest factors are also involved like growing conditions and environment (Lufu et al., 2020). A strong correlation had been recorded in weight loss and shriveling incidence by Fawole et al. (2020). Thus, lower weight loss might be the reason for having lower fruit shriveling of jamun fruit harvest from orchard-II in our experiment. Moreover, irrespective of OA-treatments and days at shelf, the fruit harvested from orchard-II exhibited overall better biochemical and antioxidative attributes of fruit as compared to fruit harvested from orchard-I. This might be due to variation in the prevailing climatic conditions such as light and temperature at both locations as these conditions determine rate of photosynthesis and supply of carbohydrates to fruit which are indispensable for all postharvest biochemical and antioxidative reactions in fruit (Sams, 1999; Tromp, 2005).

Oxalic acid (OA) treatment significantly reduced the weight loss in jamun, stored at ambient condition. Fruit weight loss depends upon various metabolic processes and respiration that result in transpiration losses from the fruit (Narayana et al., 1996). Lower metabolic activity and weight loss has been reported by the application of OA during storage at shelf and ripening (Sayyari et al., 2010). Similarly, the shriveling percentage was also reduced by postharvest application of OA in jamun. Higher metabolic activity and moisture loss is the main reason for shriveling in the postharvest life of apple fruit (Hatfield & Knee, 1988).

Moreover, the fruit SSC was lower when treated with OA that might be because of the slower conversion of starch into sugar by the application of OA. Razzaq et al. (2015) also reported lower SSC in response to OA in mango. During shelf life, SSC was slightly increased that might be due to the moisture loss of fruit which results in higher concentration of soluble solids in fruits. Similar results have also been reported by Lo'ay and El-Boray (2018) in increasing SSC contents with higher weight loss in grapes. TA was slightly affected by OA application in jamun and reduced when treated with a higher dose of OA. Maturity index (SSC/TA ratio) was found higher in orchard-I and decreased as a result of OA application which were mainly due to change in soluble solid contents of fruit. Vitamin C contents were increased as a result of 2 m*M*-OA treatment because of lower oxidation of ascorbic acid due to OA application. Being a strong antioxidant, OA suppresses the lipid peroxidation process (Kayashima & Katayama, 2002). Similar increase in vitamin C contents as a result of OA treatment has also been recorded in mango fruit (Razzaq et al., 2015).

Total phenolic contents increased with OA application at 1m*M* and slightly decreased at higher dose which indicate impact of OA depends upon the concentration. Increase in TPC with application of OA has also been reported in cherries and peaches (Cantin et al., 2009; Razavi & Hajilou, 2016). OA application has also resulted in higher levels of total antioxidants that also increased during ambient storage. Total antioxidants have a positive correlation with phenolic contents and antioxidative enzymes (Razavi & Hajilou, 2016). In current study, an increase in phenolic contents in response of OA treatment might be the reason for increasing levels of total antioxidants. Increased levels of total antioxidant activity has also been observed in other fruits like pomegranate and mango when treated with OA (Sayyari et al., 2010; Razzaq et al., 2015).

During postharvest, fruit suffer from various types of abiotic stress specially by not getting nutrients and water from the parent plant. These stresses enhance production of ROS that results in lipid peroxidation, cell wall loosening and ultimately deterioration of fruit quality (Hodges et al., 2004). SOD, CAT and POX are major antioxidative enzymes that play a significant role during various biotic and abiotic stresses. They also play a key role in extending shelf life of fruits by scavenging different Reactive Oxygen Species (ROS) produced within fruits. Activity of SOD was decreased with postharvest application of OA, while it increased the level of CAT and POX in jamun may be due to scavenging the ROS by OA and maintained a higher level of these antioxidative enzymes (Wang et al., 2009). Razavi & Hajilou, (2016) also reported a decrease in activity of SOD when OA dose increased from 1 m*M* in peach fruit whereas, activities of CAT and POX enzymes increased. Similar increase in different antioxidative enzymes has also been observed by Razzaq et al. (2015) in mangoes when OA was applied. This increase in antioxidative enzyme activity might be another reason for increasing total antioxidants in jamun when treated with OA. Total antioxidants also followed the similar decreasing trend during shelf storage as enzyme activity of SOD and CAT.

Conclusion

Harvest locations have a significant role in determination of fruit quality and in current study, the fruit harvested from orchard–I exhibited better fruit quality and antioxidants whereas shelf life was better due to less weight loss and shriveling in fruit harvested from orchard-II. Postharvest application of higher dose of OA (2 m*M*) in jamun fruit improved the shelf life due to reduction in weight loss and shriveling percentage and higher antioxidative characteristics of jamun fruit but it slightly reduced the biochemical quality as SSC and SSC/TA ratio has been reduced.

Author Contributions: I.A.R. and M.A. conceived the idea. S.U. and M.A. designed the research project, M.M.A. and U.K. executed the trial. U.N.U. analyzed the data. K.R. and G.A. wrote the manuscript. H.N.F. critically reviewed the final draft.

Acknowledgements: The authors gratefully acknowledge the financial funding of Higher Education Commission (HEC), Pakistan funded NRPU project No. 5933 entitled "Various supply chain issues and commercial potential of some minor fruits".

Conflict of Interest: The authors have no conflict of interest.

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